United States Environmental Protection Agency

LABORATORY ANIMAL PROJECT REVIEW

Please note:

1. All information in this LAPR is considered privileged and confidential by the IACUC and regulatory authorities.
2. Approved LAPRs are subject to release to the public under the Freedom of Information Act (FOIA). Do not include proprietary or classified information in the LAPR.
3. An approved LAPR is valid for three years.

LAPR Information

LAPR Title:	Assessment of disinfected drinking water and disinfection by-products for developmental and reproductive toxicity in rats.
LAPR Number:	17-06-002
Principal Investigator	Exemption 6
Author of this Document:	Exemption 6 /RTP/USEPA/US
Date Originated:	06/10/2014
LAPR Expiration Date:	06/30/2017
Agenda Date:	06/18/2014
Date Approved:	06/30/2014
Date Closed:	06/30/2017

APPROVALS

APPROVER	NAME	APPROVAL DATE	COMMENTS	
	Exemption 6 Exemption 6 Exemption 6 /RTP/USEPA/US by Exemption 6 /RTP/USEPA/US	06/30/2014	Designated Member Review	
	Exemption 6 Exemption 6 Exemption 6 RTP/USEPA/US by Exemption 6 RTP/USEPA/US	06/30/2014	DMR	

Administrative Information

1. Project Title (no abbreviations, include species):
Assessment of disinfected drinking water and disinfection by-products for developmental and reproductive toxicity in rats.

Is this a continuing study with a previously approved LAPR?
Yes
Please provide the previous LAPR# 08-03-003

2. What is the Intramural Research Protocol (IRP) number covering this project?
IRP: NHEERL-RTP/TAD/ETB/2014-001-r0
Safe and Sustainable Water Resources (SSWR) Task 2.2.D: Integrated Assessment and Reduction of Contaminant Risks

3. EPA Principal Investigator/Responsible Employee:

Principal Investigator	***Phone Number***	***Division***	***Mail Drop***
Exemption 6	Exemption 6 ***Lotus Notes Address*** Exemption 6 Exemption 6 Exemption 6 RTP/USEPA/US	TAD ***Branch*** ETB	MD

4. Alternate Contact:

Alternate Contact	***Phone Number***	***Division***	***Mail Drop***
Exemption 6	Exemption 6 ***Lotus Notes Address*** Exemption 6 Exemption 6 TP/USEPA/US	TAD ***Branch*** ETB	MD 67

SECTION A - Description of Project

1. Study objectives, presented in non-technical language such that it is understandable by non-scientific persons, including how the study addresses health protection. If this is a continuing study from a previous

LAPR, briefly justify the continuation. Please spell out all acronyms and abbreviations with their initial use.

Although disinfection of public water supplies has been a major success in decreasing disease, the disinfectant reacts with materials in the source water to produce hundreds of disinfection by-products (DBPs). Chlorinated water has been found to contain more than 600 compounds, and many DBPs remain unidentified - the known DBPs account for only approximately one-half of the mass of halogenated organic matter. Although chlorination is the disinfection method used by most US water utilities, an alternative method, chloramination is becoming increasingly common because it forms smaller amounts of the EPA-regulated DBPs. However, chloramination (i.e., disinfecting with chloramine, the result of combining chlorine plus ammonia) produces other DBPs, many of which with little, if any, toxicological data available.

Epidemiological studies, as well as toxicological studies in laboratory animals, have raised concerns regarding possible adverse health effects of DBPs. At high doses, a number of DBPs have been shown to be carcinogenic or to cause target-organ toxicity, including reproductive/developmental toxicity. However, it should be noted that many of the DBPs remain unidentified, the vast majority of the known DBPs have not been investigated toxicologically, and that little is known about the potential interactions among the DBPs. Therefore, we are using a "whole-mixture" approach to evaluate the toxicity of the complex mixtures produced by disinfection in order to better characterize the potential health risks of exposure to disinfected water. This "whole-mixture" includes unidentified contaminants along with the well-characterized DBPs, and also includes potential interactions of chemicals in mixture.

In our previous work Exemption 6Exemption 6Exemption 6) (LAPR 08-03-003), we conducted a multigenerational toxicity study to assess the reproductive effects of water that was concentrated 136 fold and then chlorinated. The 136x water was palatable for the rats, and reassuringly, the study was largely negative, particularly for prenatal mortality and pup weight; however there were slight, but significant, delays in puberty and reductions in sperm counts. The Office of Water deemed this study to be very valuable, and has asked us to conduct a similar study to compare the reproductive toxicities of chlorinated water and chloraminated water. Unlike our previous study, we plan to use a different approach to produce whole mixtures, enabling us to evaluate higher concentrations. Here, we plan to obtain freeze-dried natural organic matter (NOM) from source water. The freeze-dried NOM will then be reconstituted (with purified laboratory water) to produce water at various concentrations. The reconstituted water will be split into two "streams", one stream to be chlorinated, the other chloraminated, allowing us to conduct reproductive/developmental toxicity evaluations of chlorinated and chloraminated waters. Thus, this research will provide further insights into the potential reproductive/developmental toxicity of real-world mixtures and aid in EPA's risk assessments of chlorinated and chloraminated drinking waters, consumed by millions of people worldwide.

Using freeze-dried NOM to produce concentrated water for disinfection has several advantages over other approaches: 1) The freeze-dried NOM will be coming from a NOM repository, all produced in the same batch; thus, other researchers can obtain NOM from the same batch and link their results to our work. 2) We can produce water at higher concentrations, allowing us to more readily detect effects (using fewer animals) than with unconcentrated water. 3) We can evaluate multiple concentrations, allowing us to evaluate a dose response. 4) Volatile DBPs are not produced until disinfection (at the very end of the process); i.e., no volatile DBPs are lost during concentration.

Prior to conducting full reproductive toxicity assessments of chlorinated and chloraminated waters in Sprague-Dawley (SD) rats, we plan to conduct two preliminary studies with pregnant/lactating female rats. These studies will allow us to 1) confirm that the water preparations are palatable, and 2) assess developmental toxicity in both SD and F344 strains. Although SD rats are preferred for reproductive toxicity assays, F344 rats are more susceptible to disruptions in the endocrinology of pregnancy and eye malformations. This preliminary study will serve as a pilot for the more involved reproductive toxicity study, and provides a unique opportunity to evaluate these waters in F344 rats and provide comparisons between the two strains.

2. Scientific rationale for proposed animal use.

a. Why is the use of animals necessary?

In vivo testing is an essential part of assessing reproductive and developmental toxicity hazard in that the intact live animal is the only test system available that incorporates the complex maternal-embryonic interactions of

development.

b. Justify the species requested:

Rats have been used extensively in this field because of their size, ease of care, fecundity, and large historical database. Our work will gain insights from, and add to, the large historical database of reproductive and developmental toxicity research in this species.

3. How was it determined that this study is not unnecessary duplication?

A search of PubMed confirms that we are the only laboratory evaluating reproductive/developmental toxicity of mixtures of disinfection by-products in rodents, and is not unnecessary duplication. Search terms: reproductive toxicity, developmental toxicity, rats, mixtures, disinfection by-products, drinking water.

SECTION B - In Vivo Procedures

1. Briefly describe experimental design. Supplementary information may be attached at the end of the LAPR, but please include critical information within the body of the LAPR.

Experiment 1. Palatability study.
To assess whether or not the rats will drink the water preparations at these higher concentrations, we plan to conduct a palatability study with four treatment groups (five rats per group).

Timed-pregnant Sprague-Dawley rats will be provided water from one of the four treatments from gestation day (GD) 14 through postnatal day (PND) 21. This will allow us to evaluate whether the rats are drinking water readily during the period of peak demand (i.e., late gestation, lactation).

Freeze-dried NOM will be reconstituted with purified water (see Section D) to a volume to provide water that is 450-fold more concentrated than source water. One group will receive chlorinated concentrate, one group will receive chloraminated concentrate, and a control group will receive non-disinfected concentrate. An additional control group will receive purified water.

Control - purified water
Control - non-disinfected concentrated water (450x)
Chlorinated concentrate (450x)
Chloraminated concentrate (450x)

Maternal body weights and water consumption will be measured twice weekly.
Litters will be examined on PND 1, 5, and 21. Pups will be sexed, weighed, and examined for morphological and clinical abnormalities.

4 groups x 5 dams/group = 20 dams (SD)

Experiment 2. Developmental Toxicity Screen - strain comparison.
Timed-pregnant Sprague-Dawley and F344 rats will be provided water from one of the four treatments from gestation day (GD) 6 through postnatal day (PND) 21. This exposure period will encompass the period of embryonic organogenesis.

Treatment groups are as described for the palatability study; however, if 450x water is not palatable, the concentrations will be reduced accordingly.

Maternal body weights and water consumption will be measured twice weekly.
Litters will be examined on PND 1, 5, and 21. Pups will be sexed, weighed, and examined for morphological and clinical abnormalities.

2 strains x 4 groups x 25 dams/group = 200 dams (100 SD, 100 F344)

2. Justify the number of animals. Include explanation (e.g., biological, statistical, regulatory rationale) for the number of animals needed for each treatment group, and the overall number requested for the duration of the LAPR.

For Experiment 1 (palatability), based on our data and experience with drinking water studies, five animals per group should be adequate to identify any notable palatability issues.
4 groups x 5 dams/group = 20 dams (SD)

For Experiment 2 (strain comparison), regulatory guidelines for developmental toxicity studies recommend approximately 20 dams (i.e., with confirmed pregnancy) per treatment group. Based on historical pregnancy rates, ordering 25 timed-pregnant females per group should give us the recommended number of confirmed pregnancies.
2 strains x 4 groups x 25 dams/group = 200 dams (100 SD, 100 F344)

Overall: 20 dams (Experiment 1) + 200 dams (Experiment 2) = 220 total dams (120 SD, 100 F344)

SD rats average 12 pups per litter. If all 120 SD females bear litters, this will provide 1440 SD offspring.
F344 rats average 8 pups per litter. If all 100 F344 females bear litters, this will provide 800 F344 offspring.
1440 SD pups + 800 F344 pups = 2240 total offspring

3. State how many animals over the study period are expected to be used under the following three categories of pain/distress (USDA nomenclature as defined in the instructions): Please enter numbers only.

Categories	Adults	Offspring
C) Minimal, transient, or no pain/distress:	220	2240
D) Potential pain/distress relieved by appropriate measures:		
E) Unrelieved pain/distress:		

4. For tracking purposes, please check if this LAPR includes any of the following:

☐ Restraint (>15 Minutes) ☐ Survival surgery
☐ Food and/or water restriction (>6 Hours) ☐ Non-survival surgery

5. Category C procedures. Describe each procedure separately, include details on the following:

a. Treatments (e.g., dosages, duration of exposure, route, volume, frequency):
Exposures will be by drinking water, provided continuously, ad libitum.
Palatability study: GD 14 - PND 21
Strain comparison: GD 6 - PND 21

In Experiment 1, NOM concentrations will be equivalent to source water concentrated 450x. If Experiment 1 raises concerns regarding palatability, the concentration will be reduced accordingly in subsequent studies.

b. Survival Blood Collections (method, volume, frequency):
Experiment 2: Blood will be collected (only once per animal) on GD 10 for measurement of luteinizing hormone. Animals will be held in an acrylic restrainer for approximately 5 minutes while approximately 300 ul blood is collected from the tail vein using a butterfly needle (19G, 21G, or 23G, as appropriate for the size of the animal).
c. Testing methods (including non-stressful dietary restrictions/modifications, mild non-damaging electric shock):
Litters will be examined on PND 1, 5, and 21. Pups will be sexed, weighed, and examined for morphological and clinical abnormalities.

At necropsy of dams, uterine implantation sites will be counted.
d. Animal restraint and confinement beyond routine housing and handling. Include a description of the type of restraint device, acclimation to device, duration of restraint:
For blood collection from the tail vein, animals will be held in an acrylic restrainer for approximately 5 minutes.
e. Breeding for experimental purposes (e.g. length of pairing, number of generations):

f. Describe how animals will be monitored (e.g., frequency of observations, by whom):
Animals will be monitored by laboratory staff.
Maternal body weights and water consumption will be measured twice weekly.
Dams will be monitored several times per day for signs of parturition starting on GD 20.
In addition to thorough litter examinations on PND 1, 5, and 21 (see section B5c), cageside examinations will be done daily on weekdays to check the general condition of the litter and cases of poor maternal care.

6. Non-surgical Category D or E procedures. Describe each procedure separately, include details on the following (Also fill in Section B.9).

a. Treatments (e.g. dosages, duration of exposure, route, volume, frequency):

b. Survival Blood Collection (method, volume, frequency):

c. Testing methods:

d. Restrictions placed on the animals' basic needs (e.g., food and/or water deprivation, light cycles). Provide details regarding the length of deprivation:

e. Describe how animals will be monitored (e.g., frequency of observations, by whom):

f. Analgesia (Category D Procedures) - list drugs, dosages, route of administration and frequency:

g. If treatment-related deaths are expected, this must be thoroughly justified. Death as an endpoint is highly discouraged:

7. Surgical Category D and E procedures. Describe each procedure separately, include details on the following (Also fill in Section B.9)

a. Complete description of surgical procedure including presurgical preparation, aseptic technique, surgical closure, etc:

b. Anesthetic regimen (drugs, dosages, volume, and route of administration). The use of paralytic or neuromuscular blocking agents without anesthesia is prohibited:

c. Postoperative care (thermal support, special feeding, frequency and duration of monitoring, responsible personnel, removal of sutures/staples):

d. Post operative analgesics (drugs, dosage, and volume and route of administration, frequency):

e. Will any animals be subject to more than one major surgical survival procedures?
◯ Yes ◯ No

f. Identify any surgical procedures performed at other institutions or by vendors:

8. Humane interventions (for treatments/procedures in all categories).

a. Describe actions to be taken in the event of expected or unexpected deleterious effects from procedures or chemical exposures.
If drinking water treatments result in markedly reduced water consumption, leading to dehydration and/or deteriorating body condition, treatment may be discontinued (i.e., we will provide tap water or purified water to the animals) to allow rehydration.
We do not anticipate any maternal toxicity. However, if animals (dams or pups) show symptoms of physical injury, severe toxicity, marked dehydration, dystocia, or deteriorating body condition we will euthanize or otherwise follow AV recommendations.

b. State criteria for determining temporary or permanent removal of animals from the study.

If animals show symptoms of physical injury, severe toxicity, marked dehydration, dystocia, or deteriorating body condition we will euthanize or otherwise follow AV recommendations.

9. Alternatives to pain and distress (Category D and E Procedures only). Provide narrative regarding the sources consulted to ascertain whether acceptable alternatives exist for potentially painful/distressful procedures. Include databases searched or other sources consulted, the date of the search and years covered by the search, and key words and/or search strategy used. Assistance with searches is available through the EPA Library Staff.

SECTION C - Animal requirements

Describe the following animal requirements :

1. Indicate the number of animals required over the study period for this protocol. Please enter numbers only.

a. Animals to be purchased from a Vendor for this study: 220

b. Animals to be transferred from another LAPR:

LAPR Number that is the source of this transfer:

c. Animals to be transferred from another source:

d. Offspring produced onsite (used for data collection and/or weaned): 2240

e. TOTAL NUMBER of animals for duration of the LAPR 2460

2. Species (limited to one per LAPR): Rat(s)

3. Strain: F344, Sprague-Dawley

Describe special requirements for animals with altered physiological responses (e.g., genetically altered, aged)

none

4. Sources of animals:

Charles River Laboratories (SD rats); Harlan (F344 rats)

5. Provide room numbers where various procedures will be performed on animals:

Exemption 6

6. Will any animals be housed in areas other than the animal facility longer than 12 hours? If so, state location. Such areas require prior IACUC approval as a satellite facility before LAPR can be reviewed.

no ***Room Numbers:***

7. Describe any transportation and containment methods involved in moving animals between EPA buildings, or between EPA and other institutions (excluding any commercial shipments)

none

8. Describe any unusual housing or husbandry requirements, or acclimation requirements. Justify any treatment beginning less than 3 days after arrival.

Because of timing constraints involved with breeding and shipping, F344 animals must arrive on GD 5; treatment will begin on GD 6.

9. Describe special assistance requested of the animal contract staff, including procedures and dosing. NOTE, this request must be submitted separately to the Animal Resources Program

Office (ARPO)
none

10. Housing and Enrichment.
The IACUC encourages the use of environmental enrichment whenever possible (see IACUC website for details). Provide details on how the animals will be housed, including type of cage (e.g., solid bottom or wire screen), bedding material, number of animals per cage, and environmental enrichment. Note that housing rodents individually without environmental enrichment requires justification.
Timed-pregnant animals will be individually housed. This is necessary in order to collect individual water consumption data and also to maintain litter identity after litters are born. Both of these issues are critical to the objectives of these studies.

Heat-treated pine shavings will be used as bedding.

Enviro-dri is acceptable for use in the palatability study. However, because of concerns of endocrine disruptors, it will not be used for subsequent studies.

SECTION D - Agents Administered to Animals

1. Identify all hazardous and non-hazardous agents to be administered to living animals. For agents requiring a Health and Safety Research Protocol (HSRP), provide the title of the approved HSRP for each such agent. If no protocol is required for an agent deemed potentially hazardous (e.g. nanoparticles, recombinant DNA), describe the safety precautions to be used.
Provide maximum dosing levels and route-appropriate LD50s (where available) for each agent used for dosing.

Freeze-dried natural organic matter (NOM) will be obtained from the International Humic Substances Society, reconstituted with purified water, and provided as drinking water.
Maximum dose: 450x the concentration of source water (based on dissolved organic carbon).
LD50 data are unavailable.

Purified water (for reconstitution of NOM, and for controls) will either be deionized water, or deionized water further purified by reverse osmosis or distillation.

2. Describe any plans to administer human or animal tissues, blood or body fluids to the animals in this LAPR, and provide:

a. Information to assure that such material is pathogen-free
NA
b. A statement regarding any safety precautions necessary for handling the material.
NA

NOTE: Any unresolved health/safety questions which arise during IACUC review of this LAPR will require consultation with the Safety, Health, and Environmental Management Office.

SECTION E - Personnel Training and Experience

1. Identify all project personnel conducting animal experimentation. Specify the techniques for which they have responsibility, and their relevant training and experience. Additional personnel may be added to the table below as a group (by Division) for Category C procedures. By so doing you are giving assurance that these personnel have received all required training and are qualified to perform the Category C techniques requested.

Use this area to type in additional personnel information not available in the table drop-down lists:

Hint: *The names in the first 2 lines of the table below are filled automatically from the Principal Investigator & Alternate Contact fields. A new line will be made available when a name is selected & upon leaving the name field (i.e. tabbing or clicking in another field).*

NAME	ROLE	SPECIFIC RESPONSIBILITY	RELEVANT TRAINING
Exemption 6	Principal Investigator	Study design; litter exams, parturition exams, body weights, water bottle weights, clinical observations, tail bleeding, cervical dislocation, Category C procedures	~30 years experience. Proficient in cervical dislocation, including rats >200 g. Completed NHEERL-required training.
Exemption 6	Technical Staff	Litter exams, parturition exams, body weights, water bottle weights, clinical observations, tail bleeding, cervical dislocation, Category C procedures	~30 years experience. Proficient in cervical dislocation, including rats >200 g. Completed NHEERL-required training.
Exemption 6	Associate Principal Investigator	Study design; category C procedures	~30 years experience. Completed NHEERL-required training.
Exemption 6	Technical Staff	Litter exams, body weights, water bottle weights, clinical observations, tail bleeding	~20 years experience. Completed NHEERL-required training.
Exemption 6	Technical Staff	Litter exams, body weights, water bottle weights, clinical observations, tail bleeding	~20 years experience. Completed NHEERL-required training.
RTP-NHEERL	Tech Support	Category C Procedures	EPA IACUC Trained

SECTION F - Animal Breeding Colonies

This section pertains to the breeding of animals for maintenance of ongoing animal colonies. Do not include breeding that is part of experimentation and accountable under Section C.

Describe:

1. Estimated number of breeding pairs and liveborn per year

2. Breeding protocols and recordkeeping

3. Methods for monitoring genetic stability

4. Disposition of all offspring and retired breeders that are not used in accordance with the procedures described in this LAPR

SECTION G - Euthanasia

1. When will the animals be euthanized relative to experimental procedures?

Pups will be euthanized on PND 21 (by Animal Care Staff). If pups require euthanasia on or prior to PND 14 they will be decapitated by laboratory staff.

Dams will be euthanized on PND 21 or within in 1 week.

2. Describe the euthanasia techniques:

Method(s): Cervical dislocation
Agent(s):
Dose (mg/kg):
Volume:
Route:

Source(s) of information used to select the above agents/methods:

AVMA Guidelines for the Euthanasia of Animals: 2013 Edition
NHEERL Best Practices: Fetal and Neonatal Euthanasia

3. Provide justification and references for any euthanasia agent or method that is not consistent with recommendations of the 2007 American Veterinary Medical Association (AVMA) Guidelines for Euthanasia (e.g., cervical dislocation or decapitation without anesthesia; cervical dislocation in rodents weighing more than 200 grams).

Cervical dislocation of rats >200g: The 2013 AVMA Guidelines for the Euthanasia of Animals recommends cervical dislocation as a method of euthanasia for rats weighing <200g when performed by individuals with a demonstrated
high degree of technical proficiency. It also states that the large muscle mass in the cervical region of heavy rats makes manual cervical dislocation physically more difficult. The Guideline's 200-g weight limit is flawed for two important reasons: 1) The additional weight acquired during pregnancy or lactation has little, if any, influence on the muscle mass of the neck. (E.g., our F344 rats typically weigh 200-250 g during late pregnancy, but their nongravid weights are <180g). 2) The technique for performing cervical dislocation described by the AVMA Guidelines is appropriate for mice, but it is an inferior technique for rats. Rather than using the thumb and index finger, the preferred technique involves placing the index and middle fingers on either side of the animal's neck (from the dorsal aspect with the palm facing rostrally). Unlike the Guideline's method, this method IS appropriate for heavier animals and is NOT physically more difficult. The Principal Investigator of this project has >25 years experience performing this technique on nongravid rats weighing >350g and pregnant or lactating rats weighing >500g.

4. Describe how death is to be confirmed.

Vital organ section, Prolonged absence of breathing

SECTION H - Disposition of Used and Unused Animals

Describe the disposition of any animals remaining after project completion.

Euthanized as above, Euthanized by Animal Care Contractor, Transferred to another study

The IACUC encourages investigators to reduce the overall number of animals used at NHEERL. Would you consider transferring any unused animals from this LAPR to another approved LAPR?

● Yes ○ No

SECTION I - Assurances

1. Animals will not be used in any manner beyond that described in this application without first

obtaining formal approval of the IACUC.
2. All individuals involved in this project have access to this application, are aware of all EPA policies on animal care and use, and are appropriately trained and qualified to perform the techniques described.
3. The proposed research using animals does not unnecessarilty duplicate any previous experimentation.
4. Thorough consideration of the three "R"'s (Replacement, Reduction, Refinement) has been given, as applicable, to a. the use of animals, and b. procedures causing pain or distress (with or without analgesia/anesthesia), including death as an endpoint. The minimum number of animals required to obtain valid experimental results will be used.
5. The Attending Veterinarian has been consulted in regard to any planned experimentation involving pain or distress to animals.
6. All procedures involving hazardous agents will be conducted in accordance with practices approved by the Safety, Health, and Environmental Management Office.
7. Individuals from outside of EPA who are collaborating on this project, and who conduct related experimentation on EPA procured or bred animals in their respective Institutions, have the equivalent of a current IACUC approved LAPR at their respective Institutions.
8. The IACUC has oversight responsibilities for animal care and use, and may request consultation or feedback regarding the conduct of in vivo procedures, progress and accomplishments, and any problems encountered.

EPA Principal Investigator	Certification Signature Date
Exemption 6 by Exemption 6	06/12/2014

Submitted: 06/12/2014

Certification:

Certification by EPA Supervisor (Branch Chief or Division Director) that the project described herein has been reviewed and approved on the basis of scientific merit:

Branch Chief/Division Director	*Approval Date*	*Phone Number*	*Division*	*Mail Drop*
Exemption 6	06/12/2014 by Exemption 6 Exemption 6 Exemption 6 Exemption 6 TP/USEPA/US	Exemption 6 *Lotus Notes Address* Exemption 6 Exemption 6 Exemption 6 Exemption 6 RTP/USEPA/US	TAD *Branch* ETB	MD *Submitted to Branch Chief for Approval* 06/12/2014 02:48 PM

ATTACHMENTS

17-06-002 PI resp.pdf

Actions

First Update notification sent: 05/06/2015
Second Update notification sent: 05/27/2015
First 2nd Annual notification sent: 05/02/2016
Second 2nd Annual notification sent: 05/25/2016
1st Expiration notification sent: 05/02/2017

2nd Expiration notification sent: 06/02/2017

History Log: